REMARKS/ARGUMENTS

The Office Action of July 12, 2005 has been carefully reviewed and this response addresses the Examiner's concerns stated in the Office Action. All objections and rejections are respectfully traversed.

I STATUS OF THE CLAIMS

Claims 1-27 are still pending in the application.

Claims 1-8 and 22-27 were rejected under 35 U.S.C. 101 as being directed to non-statutory subject matter.

Claims 9-10 and 16-17 were rejected under 35 U.S.C. 101 as lacking patentable utility.

Claims 1-27 were rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enabling requirement.

Claims 1-27 were rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claims 1, 3-9, 10-14, 16, 18-22 and 24-27 were rejected under 35 U.S.C. 102(b) as being anticipated by Fox (Presentation at the 33rd BIF Conference, July 11-14 (2001)) ("Fox").

Claims 1-7 were rejected under 35 U.S.C. 102(b) as being anticipated by Loerch, (*J. Anim. Sci.*, 76(2):371-377 (1998)) ("Loerch").

Claim 2 was rejected under 35 U.S.C. 103(a) as being unpatentable over Fox in view of Tedeschi (*J. Dairy Sci.,* 83:2139-2148 (Sept. 2000)) ("Tedeschi").

Claim 8 was rejected under 35 U.S.C. 103(a) as being unpatentable over Loerch in view of Perry (J. Anim. Sci., 75:300-307 (1997)) ("Perry').

Claims 9-27 were rejected under 35 U.S.C. 103(a) as being unpatentable over Loerch in view of Pratt (U.S. 5,673,647) ("Pratt").

Support in the specification for the amendments to the claims

Claims 1, 9, 16 and 22 have been amended to include the phrase "<u>wherein (whereby) said daily feed requirement for the individual animal is provided in order to allocate feed fed to the</u>

individual animal in a pen of animals." The phrase finds support in paragraph 12 of the specification.

Claims 1, 9, 16 and 22 have also been amended to include "predicting a daily feed requirement for an individual animal, from information for the individual animal," "wherein said information comprises: initial body weight, final body weight, fat characteristics, muscle characteristics, body characteristics." These phrases find support in paragraph 35 of the specification., for example, "[e]ach individual animals' information (initial and final body weight, hip height and age, and ultrasound fat depth, rib eye area (LMA), and marbling, and body weight at the time of ultrasound) was also entered as input to the program."

Claim 22 has been amended to include the phrase "wherein means implemented in software are implemented in a computer program product comprising a computer usable medium having computer readable code embodied therein." The phrase finds support in paragraphs 37 and 42 of the specification.

II. CLAIM REJECTIONS UNDER 35 USC §101

Claims 1-8 and 22-27 were rejected under 35 U.S.C. 101 as being directed to non-statutory subject matter.

In regards to claim 1, the Examiner finds that there is not any recitation of an actual result in a form useful to one skilled in the art. Amended claim 1 states that the "daily feed requirement for the individual animal is provided in order to allocate feed fed to the individual animal in a pen of animals." Applicants in the paragraphs 4 and 5 of the specification state that "Feed costs represent 60% of the total cost incurred in the feeding of cattle. If differences in individual feed efficiency can be detected economically, this information has the potential to be used in the development of selection indexes. It is cost prohibitive to measure feed consumption on an individual animal basis in feedlots where most animals are evaluated." Therefore, there is a need and it is useful to obtain a daily feed requirement in order to allocate feed fed to an individual animal.

Further more, Applicants respectfully state that there is utility for a predicted feed requirement as evidenced by the National Academy of Science publication, *Nutrient Requirements of Beef Cattle: Seventh Revised Edition: Update 2000,* page 85, which states that "accurate estimates of feed intake are vital to predicting rate of gain;" and by the attached beef New Zealand article, available at http://www.beef.org.nz/research/breeding/breed_net_feed2.asp, which states that "An animal's expected feed intake is predicted from the test groups average feed requirements for a particular growth rate." These statements indicate that, to one skilled in the art, a predicted daily feed requirement and a predicted weight gain are useful results. Applicants respectfully state that amended claim 1 provides an actual result in a form useful to one skilled in the art and that claims 1-8, as amended, claim patentable subject matter under 35 U.S.C. § 101.

The examiner states that "a computer program is not a physical 'thing'" in rejecting claims 22 to 27, quoting MPE P 2106. Amended claim 22 restricts the means implemented in software to means implemented in a computer program product comprising a computer usable medium having computer readable code embodied therein.

Applicants respectfully restate the quote in MPEP 2106, "computer programs claimed as computer listings *per se*, i.e., the descriptions or expressions of the programs, are not physical "things." They are neither computer components nor statutory processes, as they are not "acts" being performed. Such claimed computer programs do not define any structural and functional interrelationships between the computer program and other claimed elements of a computer which permit the computer program's functionality to be realized. In contrast, a claimed computer-readable medium encoded with a computer program is a computer element which defines structural and functional interrelationships between the computer program and the rest of the computer which permit the computer program's functionality to be realized, and is thus statutory. Accordingly, it is important to distinguish claims that define descriptive material *per se* from claims that define statutory inventions." Applicants, in Paragraph 36 of the specification, state that "The one or more computer readable memories have computer readable code embodied

therein, which is capable of causing the at least one processor to execute the above described method of this invention." In other words, applicants respectfully state that the structure recited in the specification includes a computer readable medium having computer readable code embodied therein.

In *Beauregard,* Gary M. Beauregard et al. appealed the Board of Patent Appeals and Interferences decision rejecting computer program product claims as being non statutory. Since their appeal followed the *In re Lowry* decision (*In re Lowry,* 32 F. 3d 1579 (Fed. Cir. 1994).), the Commissioner stated that "computer programs embodied in a tangible medium, such as floppy diskettes, are patentable subject matter under 35 U.S.C. § 101 and must be examined under 35 U.S.C. §§ 102 and 103." *In re Beauregard,* 32 F. 3d 1583 (Fed. Cir. 1994). *In re Beauregard* is still good law. Therefore, Applicants respectfully state that claims 20 through 25, as amended, claim patentable subject matter under 35 U.S.C. § 101.

Claims 9-10 and 16-17 were rejected under 35 U.S.C. 101 as lacking patentable utility. Amended claim 9 includes the phrase "wherein said daily feed requirement and said daily weight gain for the individual animal are provided using said output means in order to allocate feed fed to the individual animal in a pen of animals." Amended claim 16 includes the phrase "whereby said daily feed requirement for the individual animal can be provided in order to allocate feed fed to the individual animal in a pen of animals." Applicants respectfully assert that, by the reasons stated above for claim 1, claims 9 and 16, as amended, have patentable utility which would be recognized by one skilled in the art.

III. CLAIM REJECTIONS UNDER 35 U.S.C. 112, FIRST PARAGRAPH

Claims 1-27 were rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enabling requirement.

The Examiner states, in the Office Action, that neither claims to the specification determined the limitation "feed requirements." One of ordinary skill in the art would know what "feed requirements" are and would know that

(6) FFG = RE/diet NEg
(7) FFM = NEm required/diet NEm
(8) Individual DM required is FFM + FFG
(9) Adjusted individual DM required = individual DM required x (total actual pen DM consumed/ pen summary of individual DM required)

Where EBW = empty body weight; EBF = empty body fat; FT = fat thickness (cm); HCW = hot carcass weight; QG = quality grade; LMA = *Longissimus dorsi* muscle area (cm^2); SBW = shrunk body weight; AFSBW = weight at 28% body fat; EQSBW or EQEBW = shrunk or empty body weight equivalent to the standard reference animal; EBG = empty body gain; RE = retained energy, Mcal/day; FFM = feed for maintenance; FFG = feed for gain; and DM = dry matter.
As given in paragraph 27 of the specification provides a method for computing the feed requirements. Also, Applicants submit, as examples,
http://www.agric.nsw.gov.au/reader/beefnutrition/dai12.htm or
http://www.oznet.ksu.edu/library/agec2/samplers/MF264.asp or
http://www.maf.govt.nz/mafnet/rural-nz/sustainable-resource-use/best-management-practices/reassessment-of-the-stock-management-system/re-assessment-of-stock-unit-system05.htm
that use of the term "feed requirements" and the same terminology as used in the specification, the examples being from three different countries and two continents.

While the Examiner states that the skill of those in the art of bioinformatics is high, the art here is an animal husbandry, the feeding of cattle and matter as studied in the school of agricultural science.

The Examiner, in page 7 of the Office Action, cannot find items in the specification for protecting feeding requirements taken into account in genotype type and measuring actual gain.

Measuring actual gain is something that one of less than ordinary skill in the art would know how to do (the skill required is one of your average 4- H. club member).

The Cornell Net Carbohydrate and Protein System is well known in the art as evidenced by http://www.dasc.vt.edu/extension/nutritioncc/polan98.pdf where it is stated that the CNCPS includes maintenance requirements sensitive to animal and environmental conditions, or http://www.soyasa.org/documents/1/Firkins_P_Soy_Protein_in_Dairy_Jun04.pdf states that probably the most widely used model for the rumen process in the US is the Cornell net carbohydrate and protein system.

The specification states, in paragraph 24, that "net energy for gain, NEg, and net energy for maintenance, NEm, concentration of the diet are determined taking into account genotype and environmental differences (step 20, Fig. 1a; step 15, Fig. 1b). Environmental differences include variables such as outside temperature, internal and external insulation, wind, and hair coat depth and condition as well as activity. In one embodiment, the net energy for gain, NEg, and Net energy for maintenance, NEm, concentration of the diet are determined utilizing the Cornell Net Carbohydrate and Protein System (CNCPS) (see Fox, D. G., T. P. Tylutki, M. E. Van Amburgh, L. E. Chase, A. N. Pell, T. R. Overton, L. O. Tedeschi, C. N. Rasmussen, and V. M. Durbal, The Net Carbohydrate and Protein System for evaluating herd nutrition and nutrient excretion: Model documentation. Version 5.0, Mimeo No. 213, Animal Science Dept., Cornell University, Ithaca, NY, 2003)." One of ordinary skill in the art, utilizing an extremely well-known model, the CNCPS, would know how to perform the calculations and include both genotype and environmental considerations.

Furthermore, in U.S. Provisional Application 60/396,361 entitled "Computer Model for Individual Cattle Management," filed on July 17, 2002, which was incorporated by reference in the applicants' specification, in p.5, second paragraph, entitled "*Accounting for Differences in Requirements for Maintenance,*"

states that:

> The model used for this purpose is described by Fox and Tylutki (Fox, D. G. and T. P. Tylutki. 1998. Accounting for the effects of environment on the nutrient requirements of dairy cattle. J. Dairy Sci. 81:3085-3095.) The effects of breed type are accounted for by adjusting the base NE_m requirement of 77 kcal/kg$^{0.75}$ of BW (MBW, metabolic body weight) for *Bos indicus* and dairy types (-10 and +20% compared to *Bos taurus*). The effects of previous nutrition are accounted for by

> relating body condition score to NE_m requirement. On a 1 to 9 scale, maintenance requirement is reduced 5% for each condition score below 5 and is increased 5% for each score above 5. The effects of acclimatization are accounted for by adjusting for previous month's average temperature (ranges from 70 kcal/kg MBW at 30 °C to 105 kcal/kg MBW at -20 °C). This adjustment is continuous, with no effect at 20 °C. Current environmental effects are accounted for by computing heat lost vs heat produced, based on current temperature, internal and external insulation, wind, and hair coat depth and condition. This becomes important when the animal is below the computed lower critical temperature, and can range from no effect at 20 °C to twice as high (thin, dirty hide at -12 °C and 1 mph wind),

thereby providing further enablement of taking into account genotype and environmental considerations.

The Examiner states that the claims do not recite any models or algorithms and the example in the specification indicates additional steps are required. As it has been stated many times by the Federal Circuit Court of Appeals, "claims claim, specifications teach."

In paragraphs 24 through 31 of the specification, the invention is described and examples are presented with sufficient detail that one skilled in the art, the art being animal husbandry, would know how to make and use the invention. Another example is presented in U.S. Provisional Application 60/396,361 entitled "Computer Model for Individual Cattle Management," filed on July 17, 2002.

The description in the specification refers to a model, the CNCPS, that is well-known in the art and is disclosed "in a way that one skilled in the art will understand" (MPEP 2181, quoting *Atmel Corp v. Information Storage Devices, Inc.* 198 F. 3d 1374, (Fed. Cir. 1999) where the Federal Circuit concluded that the title of the article in the specification may be sufficient to indicate the one skilled in the art the precise structure). The formulas and steps in Table 1 and Table 3 and paragraphs 25-34 of the specification would enable one of ordinary skill in the art to practice the invention.

The Examiner states, in page 7 of the office action that in the example provided in the specification requires additional steps/information that are not recited in the claims. The

specification states, in paragraph 35, that the individual animals' information is provided as input to the program. The inputs are not part of the inventive contribution and the information listed in paragraph 35 is information usually collected in the performance of the art (as would be done by one of ordinary skill in the art).

Therefore, Applicants respectfully assert that claim 1-27, as amended, comply with the enabling requirement of 35 U.S.C. 112, first paragraph.

IV. CLAIM REJECTIONS UNDER 35 U.S.C. 112, SECOND PARAGRAPH

Claims 1-27 were rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Amended claims 1, 9, 16 and 22 claim the predicting of a daily weight gain animal <u>from the daily feed requirement,</u> thereby expressing a relationship between the steps. Amended claims 1, 9, 16 and 22 also include the phrase "<u>wherein (whereby) said daily feed requirement for the individual animal is provided in order to allocate feed fed to the individual animal in a pen of animals,</u>" relating to steps to the individual feeding of cattle.

Furthermore, in regards to the preamble, "The determination of whether preamble recitations are structural limitations or mere statements of purpose or use "can be resolved only on review of the entirety of the [record] to gain an understanding of what the inventors actually invented and intended to encompass by the claim." *Corning Glass Works,* 868 F.2d at 1257, 9 USPQ2d at 1966. If the body of a claim fully and intrinsically sets forth all of the limitations of the claimed invention, and the preamble merely states, for example, the purpose or intended use of the invention, rather than any distinct definition of any of the claimed invention's limitations, then the preamble is not considered a limitation and is of no significance to claim construction. *Pitney Bowes, Inc. v. Hewlett-Packard Co.*, 182 F.3d 1298, 1305, 51 USPQ2d 1161, 1165 (Fed. Cir. 1999)." MPEP 2111.02

The preamble in claim 1 merely states the intended use of invention and therefore should not be consider a limitation.

The term "animal," as used in this invention, is defined in the specification in paragraph 21 as referring "to feedlot steers or heifers, or herd replacement bulls or heifers." The applicant can act as his own lexicographer (MPEP 2111 .01).

The term "daily feed requirements," as stated above, is a known term in the art and has the ordinary and customary meaning attributed to them by those of ordinary skill in the art, as stated above.

The term "Environmental differences" is defined in the specification (in paragraph 24) as including "variables such as outside temperature, internal and external insulation, wind, and hair coat depth and condition as well as activity."

The term "genotype" is defined in the specification, in paragraph 21, as including breed type, body size, stage and rate of growth. One skilled in the art of animal husbandry would not interpret genotype as genomic mutations, SNPs (one skilled in the art of animal husbandry would not know what an SNP is), or polymorphism. The use of the term "genotype" as defined by the Applicants in the specification is in agreement with the use of that term in the art, the art being animal husbandry or animal science, as can be seen, for example, from "The Effect Of Cattle Genotype On Texture Of Selected Muscles During Postmortem Aging", found at http://www.ejpau.media.pl/issues/volume8/issue3/index.html, from http://www.fao.org/docrep/003/t0413e/T0413E05.htm, and from http://www.ams.usda.gov/lsg/certprog/schedules/chblaspec.htm , copies of which are included in the Appendix.

The term "actual gain," as stated above, is well understood in the art, and one of less than ordinary skill in the art would know how to obtain the actual gain (a cowhand or a junior high school 4H club member would know how to obtain the actual gain).

The term "individual animal characteristics," is used in the specification to include hot carcass weight; QG = quality grade; LMA = *Longissimus dorsi* muscle area (cm^2) (paragraph 25) and initial and final body weight, hip height and age, and ultrasound fat depth, rib eye area (LMA), and marbling, and body weight at the time of ultrasound (paragraph 35).

The term "adjusted shrink body weight" is a known term in the art but it is defined in the specification as “adjusted to 28% fat” (paragraph 21). (For uses in the art see, for example, www.clemson.edu/avs/extension/Jul02.pdf, and jas.fass.org/cgi/content/full/80/7/1791.)

The term "taking into account" has its plain and usual meaning of "taking into consideration" or "including the effects of."

The term "performance information" is defined in claims 4, 12, 19 and 25 by the wherein clause “wherein the performance information comprises feed intake, and feed efficiency,” which was present in the claims as originally filed.

Therefore, Applicants respectfully assert that claim 1-27, as amended, comply with the definiteness requirement of 35 U.S.C. 112, second paragraph.

V. CLAIM REJECTIONS UNDER 35 U.S.C. 102(b)

Claims 1, 3-9, 10-14, 16, 18-22 and 24-27 were rejected under 35 U.S.C. 102(b) as being anticipated by Fox (Presentation at the 33rd BIF Conference, July 11-14 (2001)) (“Fox”).

Enclosed is a 35 CFR 1.132 declaration by Professor Danny Fox, one of the Applicants and the presenter at the 33rd BIF Conference, in which Professor Fox states that no handouts were given at the conference, the paper was written after the oral presentation and that the proceedings were distributed at least two weeks after the conference. Based on the 35 CFR 1.132 declaration by Professor Fox, the publication utilized by the Examiner was not available to the public or in a

printed publication one year prior to the filing date of the provisional application 60/396,361 (filed on July 17, 2001) that establishes the priority date of the present application. Therefore, applicants respectfully assert that the paper published in the proceedings of the 33rd BIF Conference, July 11-14 (2001) is not prior art under 35 U.S.C. 102(b).

Furthermore, the paper published in the proceedings of the 33rd BIF Conference, July 11-14 (2001) does not disclose all of the Applicants' claimed invention (as can be seen by the absence of an equivalent of Table 3 in the Applicants' specification from the paper published in the proceedings of the 33rd BIF Conference) and therefore does not disclose or enable all the limitations of the Applicants' claimed invention. and would not anticipate the Applicants' claimed invention.

Claims 1-7 were rejected under 35 U.S.C. 102(b) as being anticipated by Loerch et al, (J. Anim. Sci., 76(2):371-377 (1998)) ("Loerch").

Regarding Loerch, Loerch et al "conducted two experiments to determine the effects of various intake restriction strategies on performance and carcass characteristics of feedlot cattle" and do not predict performance of individual animals on a daily basis. In the Applicants' claimed invention, daily feed requirements for an individual animal and a daily weight gain for an individual animal are predicted. Loerch et al. report their experimental findings in terms of an average daily feed intake and an average daily gain for 105 steer calves of the same genotype (crossbred - Continental x English) (see Loerch, p. 373, second column, last paragraph, also all tables provide one number for all 105 calves).

Comparisons of the limitations of claim 1 and the disclosures given by Loerch are given in the table below.

Claim 1 - limitations	Loerch
predicting a daily feed requirement for an individual animal, from characteristics for the individual animal, taking into account genotype, diet, and environmental differences	Tables 1-4 - one (1) prediction over the entire growing period for one genotype (crossbred - Continental x English) - not daily, no genotype differences, no environmental differences in the prediction.
predicting a daily weight gain of the individual animal from said daily feed requirement;	Tables 1-4 - one (1) prediction over the entire growing period for one genotype (crossbred - Continental x English) - not daily, no genotype differences, no environmental differences in the prediction.

Regarding claims 2-7, Loerch in pages 372, 373, 374, Table 3 and table, provides measured results of experiments (see col. 1, p.373 3rd paragraph. "Steer performance data for Exp. 1 are given in Table 3," p. 375, col. 2, paragraph 2, " Steer performance data for Exp. 2 are given in Table 5."). Applicants respectfully assert that while one skilled in the art would know how to measure the measured quantities, Applicants' claimed invention relates to predicting for an individual animal. Applicants also respectfully assert that disclosures by Loerch which were compared to the limitations of claims 2-7 were all related to experimental measurements and conditions and not to predictions.

Since Loerch does not teach or disclose, explicitly or implicitly, "predicting a daily feed requirement for an individual animal taking into account genotype, diet, and environmental differences" or "predicting a daily weight gain of the individual animal from the daily feed requirement," Applicants respectfully assert that Loerch does not teach or disclose any of the limitations of the Applicants' claimed invention.

To anticipate a claim a reference must teach every element of the claim. (MPEP § 2131). "A claim is anticipated only if each and every element as set forth in the claim is found, either

expressly or inherently described, in a single prior art reference." *Verdegaal Bros. v. Union Oil Co. of California*, 814 F.2d 628, 631, 2 USPQ2d 1051, 1053 (Fed. Cir. 1987). Since Loerch does not teach or disclose, explicitly or implicitly, at least one limitation of the Applicants' claimed invention, applicants respectfully assert that claims 1-7 are not anticipated by Loerch.

VI. CLAIM REJECTIONS UNDER 35 U.S.C. 103(a)

Claim 2 was rejected under 35 U.S.C. 103(a) as being unpatentable over Fox in view of Tedeschi (J. Dairy Sci., 83:2139-2148 (Sept. 2000)) ("Tedeschi").

As stated above, the paper published by Fox in the proceedings of the 33rd BIF Conference, July 11-14 (2001) is not prior art under 102(b), and even if it were, it would not anticipate the Applicants' claimed invention since it is lacking at least one limitation of the Applicants' claimed invention.

Applicants also respectfully state that Applicants have not claimed that the CNCPS comprises an iterative step and that neither have Tedeschi et al. (J. Dairy Sci., 83:2139-2148 (Sept. 2000)). In one embodiment of the Applicants' claimed invention, the CNCPS is used to provide the net energy for gain and the net energy for maintenance (see paragraph 24 of the specification). In Tedeschi et al. (J. Dairy Sci., 83:2139-2148 (Sept. 2000)), Tedeschi et al. iterate over the amount of feed components (such as starch, fat, protein B1) in order to obtain an optimal diet according to an optimization criterion (see p. 2140 in Tedeschi et al., see also table 2, Figure 1, on the outputs in Figures 5 and 6 in Tedeschi et al.). Tedeschi et al. do not teach or disclose or suggest predicting a daily weight gain. Tedeschi et al. do not teach or disclose or suggest predicting a daily feed requirement for an individual animal or a daily weight gain for individual animal.

Therefore, Applicants state that the paper published by Fox in the proceedings of the 33rd BIF Conference, July 11-14 (2001) is not prior art under 102(b) and unavailable for combination with Tedeschi et al. and, even if it were, since neither of the references nor the combination teach

or suggest all the limitations of claim 2, a prima facie case of obviousness would not have been established.

Claim 8 was rejected under 35 U.S.C. 103(a) as being unpatentable over Loerch in view of Perry (J. Anim. Sci., 75:300-307 (1997)) ("Perry").

As stated above, Since Loerch does not teach or disclose, explicitly or implicitly, "predicting a daily feed requirement for an individual animal taking into account genotype, diet, and environmental differences" or "predicting a daily weight gain of the individual animal from the daily feed requirement." Applicants respectfully state that "Perry" does not teach or disclose, explicitly or implicitly, "predicting a daily feed requirement for an individual animal taking into account genotype, diet, and environmental differences" or "predicting a daily weight gain of the individual animal from the daily feed requirement." Since claim 8 is dependent on claim 1, and neither Loerch nor "Perry," individually or in combination, teach or suggest any of the limitations of claim 1, Applicants respectfully state that that Loerch in view of "Perry" does not teach or suggest at least one distinct limitation of claim 8.

"To establish a *prima facie* case of obviousness, three basic criteria must be met. First, there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references when combined) must teach or suggest all the claim limitations." MPEP 2143

Therefore, Applicants respectfully state that a prima facie case obviousness has not been established, and that claim 8 is patentable over Loerch in view of "Perry."

Claims 9-27 were rejected under 35 U.S.C. 103(a) as being unpatentable over Loerch in view of Pratt (U.S. 5,673,647) ("Pratt").

As stated above, Loerch does not teach or disclose, explicitly or implicitly, "predicting a daily feed requirement for an individual animal taking into account genotype, diet, and environmental differences" or "predicting a daily weight gain of the individual animal from the daily feed requirement." Pratt teaches obtaining an average daily gain (see col. 15, lines 64-65, the '647 patent, see also Figure 25). Pratt does not teach or disclose explicitly or implicitly, "predicting a daily feed requirement for an individual animal taking into account genotype, diet, and environmental differences" or "predicting a daily weight gain of the individual animal from the daily feed requirement," or a computer usable medium having computer readable code which that causes a processor to execute those two steps, a limitation of independent claim 9 and of independent claim 16. Neither Loerch nor Pratt, teach or disclose explicitly or implicitly, "means for predicting a daily feed requirement for an individual animal taking into account genotype, diet, and environmental differences" or "means for predicting a daily weight gain of the individual animal from the daily feed requirement," which are limitations of independent claim 22. Therefore, neither Loerch nor Pratt, individually or in combination, teach us suggest at least one limitation of claim 9, at least one limitation of claim 16, or at least one limitation of claim 22.

Since claims 10-15 are dependent on claim 9, claims 17-21 are dependent on claim 16, claims 23-127 are dependent on claim 22 and neither Loerch nor Pratt, individually or in combination, teach us suggest at least one limitation of claim 9, at least one limitation of claim 16, or at least one limitation of claim 22, Applicants respectfully state that that Loerch in view of Pratt does not teach or suggest at least one distinct limitation of claims 9-27.

Therefore, Applicants respectfully state that a prima facie case obviousness has not been established, and that claims 9-27 are patentable over Loerch in view of "Pratt."

VII. CONCLUSION

In conclusion, in view of the above remarks, Applicants respectfully assert that the claims in this application are now in condition for allowance and respectfully request the Examiner find claims 1-27 allowable over the prior art and pass this case to issue.

Although no additional fees are anticipated, the Commissioner for Patents is authorized to charge additional fees or credit overpayment to Deposit Account No. 03-2410 (Order No. 10845-147).

The following information is presented in the event that a call may be deemed desirable by the Examiner:

ORLANDO LOPEZ (617) 854-4000.

Respectfully submitted,
Luis O. Tedeschi et al., Applicants,

Date: October 12, 2005

By:
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THE ROLE AND MECHANISMS OF GENETIC IMPROVEMENT IN PRODUCTION SYSTEMS CONSTRAINED BY NUTRITIONAL AND ENVIRONMENTAL FACTORS

By

Ola Syrstad

INTRODUCTION

One of the many constraints on milk production in the tropics is the poor genetic potential of the indigenous animals. Tropical cattle are mostly of zebu (Bos indicus) type. These cattle are well adapted to the conditions prevailing in the tropics. Natural selection over hundreds of generations has provided them with a high degree of heat tolerance, some resistance to many tropical diseases and the ability to survive long periods of feed and water shortage. However, their dairy potential is poor; they have low milk yield, they are late maturing and usually do not let down milk unless stimulated by the sucking of the calf.

Genetic improvement alone might not result in drastic increases of milk production in the tropics, but it is a prerequisite for such increases. Genetically more productive animals are also the best incentive to improved feeding and management.

GENOTYPE AND ENVIRONMENT

The performance of an animal is the result of the joint action of its genotype and the non-genetic effects to which it is exposed. The non-genetic factors are often collectively termed the "environment".

The genotype is often conceived as a frame which restricts the performance to a given level. Below this level, the performance is determined by the environment. This concept is visualized in Figure la. Two genotypes, A and B, are considered. The superiority of the better genotype, A, is expressed only if the environment is more favourable than that which is necessary to exploit fully the potential of the poorer genotype, B. When the environment is worse than this, both genotypes would perform similarly and genetic improvement beyond the level of B would be of no use. According to this model both genotype and environment can act as bottlenecks which restrict performance. Although this might seem reasonable, the available evidence indicates that the concept is in general not providing an appropriate description of the interaction between genotype and environment.

The model illustrated in Figure 1b is much more likely to be correct in most cases. Here the superiority of the better genotype, A, is realized, regardless of the environmental conditions, but the difference between the two genotypes increases as the environment improves. This means that genotype A responds more to improved conditions than genotype B (indicated by the steeper slope of the line), but the genetic difference is expressed also under poor conditions. Most research on genotype - environment interaction in dairy cattle supports this model (review by Syrstad, 1976). In studies on field data, progenies of various bulls have been found to rank very similarly over a wide range of production levels. The same was true when progenies of the

same bull in Mexico and U.S./Canada were considered (McDowell et al., 1975). A recent review of dairy cattle crossbreeding in the tropics (Syrstad, 1989) suggests that the relative merits of two genetic groups (1/2 vs. 3/4 exotic inheritance) is independent of production level.

Figure 1. Different models of genotype x environment interaction. For explanation, see text.

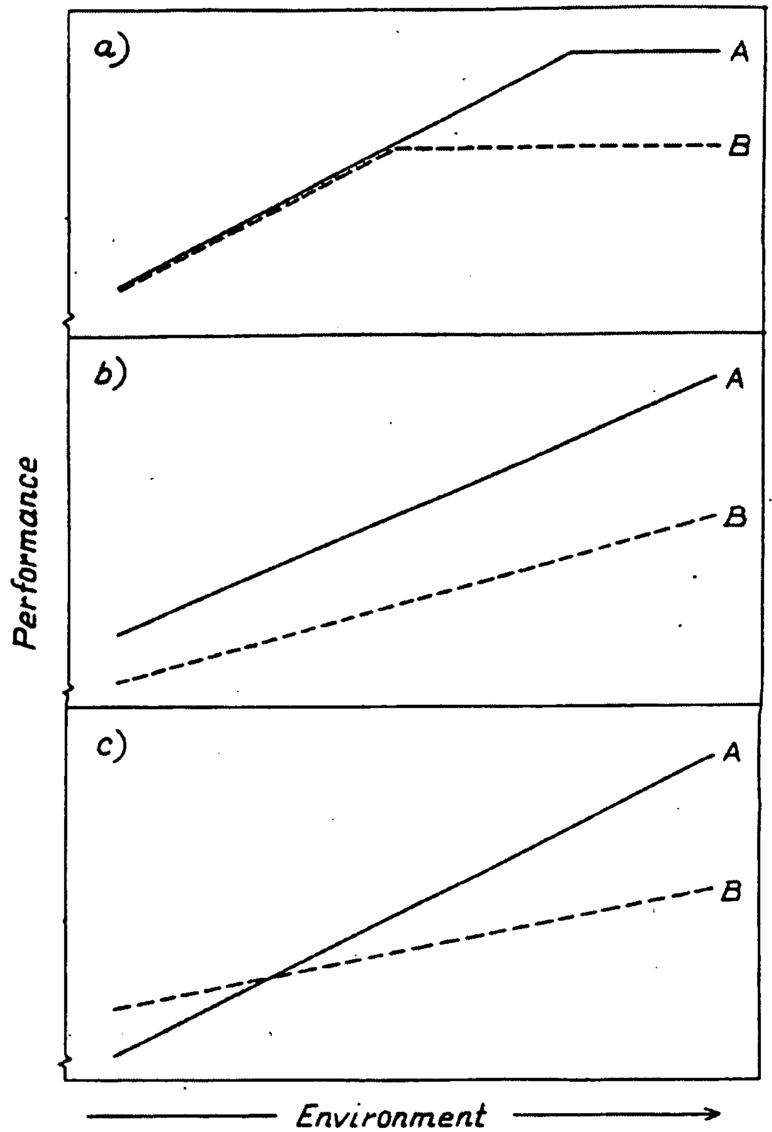

Figure 1c describes a situation in which the different responses of the two genotypes to environmental improvement results in reversed ranking. Genotype A is the better under good conditions, while B is superior when the environment falls below a given level. This might occur in cases when the environment varies over a very wide range. An example from beef cattle is presented by Hearnshaw & Barlow (1982). Crosses of Hereford with Friesian, Simmental and Brahman (American Zebu) were compared under good, intermediate and poor pasture conditions. The Friesian and Simmental crosses were the best on good pasture, while Brahman crosses were superior on poor pasture. In dairy cattle, Buvanendran & Petersen (1980) found almost no relationship between the performance of daughters of the same bull in Denmark and Sri Lanka. However, the number of daughters in Sri Lanka was small, and the lack of association might be incidental.

Model (b) suggests that the best breeding strategy is to select breeding animals on their performance in a good environment, as this is when the genetic differences between animals is most clearly expressed. But this would be dangerous if model (c) should be correct. The safest, and usually the most efficient, approach is to base selection on the merits of the animals as expressed under environmental conditions similar to those which their progenies will be exposed to.

METHODS FOR GENETIC IMPROVEMENT

a) Selection within the local population

Cattle indigenous to the tropics have, except in very few cases, been subjected to only little artificial selection for increased milk production. In view of the impressive results achieved by selection in many temperate dairy breeds there should be good prospects for improving the dairy potential of tropical cattle by the same method.

Genetic improvement per generation from selection depends on the variability of the traits considered, their heritability (i.e. the proportion of total variation which can be ascribed to genetic differences), and the intensity of selection. Variability, in terms of the coefficient of variation, is usually greater in tropical than in temperate cattle, but the variation in actual units is less. Studies of heritability based on sufficiently large amounts of data are few, but estimates reported fall within the same range as those from temperate countries. Intensity of selection is restricted by the reproductive rate, and is further reduced by early mortality, which often is high under tropical conditions.

Many dairy cattle breeding programmes claim a genetic improvement in milk yield of one to two percent per year. Of this improvement, 60 to 70 percent is derived from the selection of bulls on the basis of the performance of their daughters (progeny testing). This is achieved by a combination of accurate progeny testing (i.e. many daughters per bull) and intensive selection (many bulls tested per year). These conditions can be fulfilled only in large populations, comprising tens (if not hundreds) of thousands of females, artificial insemination, and widespread milk recording.

In most tropical countries such populations do not exist, and are not likely to be available in the foreseeable future. Instead a breeding programme might have to be established in a single herd or a few cooperating herds. In order to make progeny testing worthwhile, even strictly on genetic grounds, several hundred females would be needed. Still, the high costs involved might not make such a programme attractive. But if the herd serves as a nucleus herd, also providing bulls for breeding outside the herd, the benefit of genetic progress will in turn be transmitted to a much larger number of animals, and it might be justified to maximize genetic progress in spite of high costs. Thus a rather small breeding scheme can have tremendous impact if organized and operated properly.

b) Introduction of improved tropical breeds

Some breeds of tropical cattle, e.g. Sahiwal and Red Sindhi, have been selected for increased milk yield over a long time and have reached a much higher dairy potential than most cattle in the tropics. This is a genetic resource which should be exploited for upgrading of unimproved stock. After a few generations of back-crossing to bulls of the improved breed, the inheritance of the local cattle has been almost completely replaced by the improved inheritance. The risk of losing adaptability to local conditions by this method is small, a breed like Sahiwal has shown to adapt well to conditions in four different continents. An improvement which would require ten generations of intensive selection could be obtained in two or three generations of upgrading with an improved breed. Unfortunately the number of animals of improved tropical breeds is small, and breeding stock of high quality are not easily available.

c) Introduction of temperate breeds

Reports on the high milk yields in some temperate countries have spread the belief that the importation of European-type dairy breeds is the solution to the problem of low production levels in the tropics. In some cases introduction of temperate breeds has been successful but much more often the experience has been disappointing and sometimes almost disastrous. Diseases, high mortality rates and low fertility have been frequent problems among the imported animals and their progenies, and animals which have survived have failed to reach the expected production levels. Offspring born in the tropical country have often produced much less than their dams, which were imported as heifers. The lack of adaptation to tropical conditions has been obvious. On the basis of experience up to this time, purebred European-type dairy cattle can be recommended in the tropics only if climatic stress is moderate, health services are easily available and reasonably good feeding is practised.

d) Crossbreeding with European type cattle

Crossbreeding of tropical cattle with cattle of European-type breeds has occurred for more than one hundred years, and a large number of reports has been published. In most cases, females of local stock have been mated to bulls of the imported breed or by the use of imported semen.

In almost all cases, crossbreeding with a European breed led to a dramatic increase in milk yield in the first crossbred generation (F1), compared with the local stock. The crossbred females calved at a much younger age than native animals, produced two to three times more milk and had longer lactations, shorter dry periods and shorter calving intervals. Mortality and susceptibility to disease were only slightly higher than in native cattle.

These favourable results were, naturally, ascribed to the superiority of the exotic inheritance, and it was tempting to introduce more of it by backcrossing to exotic bulls. But the expected further improvement did often not occur and in many cases a decline in performance was observed. Problems of high mortality and reduced fertility increased as the level of exotic inheritance increased towards 100 per cent.

When it had been found that upgrading towards the European breed was not advisable under most conditions, the next step was to try to stabilize the level of exotic inheritance by mating F1 males and females together. But again the results were often disappointing. In almost all projects the performance of the second half-bred generation, F2, has been much below that of F1. Age at first calving and calving intervals have increased considerably and milk yield has dropped by up to 30 per cent.

A summary of results from 54 sets of data reported from cross-breeding experiments in various regions of the tropics is presented in Table 1. The good performance of the first crossbred generation (F1) and the deterioration in the next generation (F2) are clearly demonstrated. The most obvious explanation is the presence of hybrid vigour (heterosis); this effect is maximized in the F1 but half of it is expected to disappear in the F2 and forward generations. In addition other genetic mechanisms might also be involved.

The great effect of hybrid vigour in crosses of zebu x European-type cattle might be expected because of the wide genetic distance between the two types (Cunningham and Syrstad, 1987). Furthermore it has been suggested that hybrid vigour is more important under stressful than under favourable environmental conditions (review by Barlow, 1981). The breeding strategy for dairy cattle in the tropics should therefore also aim at exploiting hybrid vigour. Exactly how this can be done under various conditions is still a question for discussion, and more research is needed.

Table 1. Performance of zebu cattle, European type cattle, and their crosses in the tropics. Summary of 54 sets of data. Source: Syrstad (1988).

Proportion European cattle	Age at first calving, months	Milk yield, kg	Calving interval, days
0 (i.e. zebu)	43.6	1052	459
1/8	40.1	1371	450
1/	37.5	1310	435
3/8	36.1	1553	435
1/2 (F1)	32.4	2039	429
5/8	33.8	1984	432
3/4	33,9	2091	450
7/8	34.4	2086	459
1 (i.e. European)	31.6	2162	460
1/2 (F2, from F1xF1)	33.7	1523	449

Table 2. Comparison of F1 and backcrosses (1/2 and 3/4 European inheritance) at low, intermediate and high production levels. Summary of 30 sets of data. Source: Syrstad (1989).

Production level	Average milk yield, kg	
	F1	Backcrosses
Low (<2000 kg)	1487	1605
Intermediate (2000–2405 kg)	2175	2218
High (>2405 kg)	2798	2698

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LIVE ANIMAL SPECIFICATION
AMERICAN HEREFORD ASSOCIATION'S
SPECIFICATION FOR CHARACTERISTICS OF CATTLE ELIGIBLE FOR THE AMERICAN HEREFORD ASSOCIATION CERTIFIED HEREFORD BEEF PROGRAM
DECEMBER 2003

I. SCOPE

This specification is for use by representatives of the American Hereford Association (AHA) and its designates to assure that cattle presented for Certified Hereford Beef (CHB) program certification by the Meat Grading and Certification Branch (MGCB) of USDA meet the live animal requirements of the CHB program. The purpose of this specification is to define visual indicators of cattle that are at least 50% Hereford and 100% English breed type to the exclusion of dairy, continental, and bos indicus genetic type cattle.

II. REQUIREMENTS

(a) Phenotype

- Cattle offered for AHA qualification must have a predominately (51%) white face. Cattle must exhibit white markings over the jaw, forehead and muzzle. Cattle must have white markings on all three locations or they are unacceptable.
- Cattle must exhibit some or all of the traditional markings of a Hereford or Hereford/English crossbred animal such as white on the face and neck. White markings on any part of the hip, shoulder or side of the body such as spots, stripes, or belts are not acceptable.
- Cattle offered for qualification other than solid red, solid black, or roan with a predominately white face, such as yellow, gray, brown, brindle, or smoke are ineligible.
- Steers and heifers presented for AHA qualification must be of beef type breeding. Animals must express some evidence of muscle bulge. Although not limited to, this bulge is best observed in the large muscles of the round.
- An animal expressing dairy type breeding (dairy-type head, prominent "hook" bones, large "barrel-like" body, etc.) typically has muscles in the round which are semi-flat to concave or "dished" in appearance. Such animals are not acceptable.
- Cattle offered for qualification shall not have an excessive hump on the withers (protruding above the top line), excessively long ears (ears that project downward from the head) or an elongated head indicative of bos indicus genetics. The hump on the withers should not be confused with a crest on the neck, which is normal for many thick muscled cattle, particularly males.
- To qualify, cattle may be either horned or polled. Bulls, bullocks, or cows are not acceptable.

(b) Genotype

In lieu of the aforementioned phenotypic requirements regarding color and traditional Hereford markings, qualification can be based upon a signed affidavit provided by the cattle breeder that verifies the cattle are at least genetically ½ Hereford breeding and 100% British bred (Angus, Red Angus, Shorthorn, Hereford, or South Devon). All genotype cattle must be the direct offspring of a registered Hereford bull or cow. This affidavit must be submitted to a representative of the AHA prior to feedlot qualification. Cattle qualifying by genotype documentation must be presented for slaughter as a unique lot separated from all phenotype qualified cattle and non-qualifying cattle and designated as "Genotype" cattle on the kill sheet. Genotype qualified cattle may not change ownership after they have been assigned an official AHA lot number and have been approved for slaughter.

CHB Harvest Monitoring Procedures

At the time of slaughter the approved personnel shall determine those animals that meet the CHB Live Animal Specifications and identify the carcasses of those animals with a "C" stamp. With regard to cattle that are genetically identified, the identity of the qualifying lot must be maintained until the "C" stamp is applied to the carcass.

USDA Meat Grading and Certification Branch representatives shall conduct unannounced, random checks of the AHA approved personnel to monitor compliance with these procedures. AHA approved personnel shall be designated with a program specified helmet sticker. MGCB agents shall observe the work of the approved plant employee for a period of two to five minutes. During that period defects shall be recorded for any animal which is identified with a "C" stamp, but which fails to meet one or more of the specified requirements.

Frequency of observations shall be determined according to the rate of slaughter during a designated work shift as shown in Table 1. Any observations shown in Table II as defects shall be recorded during each monitoring period.

TABLE I. MONITORING FREQUENCY

Slaughter Rate Number of cattle/shift	Frequency 1/ Number of monitoring periods 2/
Up to 500	2
501 – 1,000	4
1,001 – 1,500	6
1,501 – 2,000	8
Over 2,000	10

1/ The MGCB agent shall randomly determine the times of actual observation before the beginning of the slaughter shift.

2/ The MGCB agent shall observe the work of the approved plant employee for a period of not less than 2 nor more than 5 minutes.

Defect Categories Major	Minor	Defects
101		30 percent or less of the face is white
	201	31 percent to 50 percent of the face is white
102		Cattle with 50 percent white face are other than solid red, solid black, or roan
	202	Cattle with 50 percent white face are light red and do not show additional Hereford markings
103		Cattle with markings on the hip, shoulder, or side of the body including spots, stripes, or belts.
104		Presence of two or more of the following bos indicus indicators: excessive hump, excessive ears, or bos indicus head
	204	Presence of one of the following bos indicus indicators: excessive hump, excessive ears, or bos indicus head
105		Animal qualified which has predominant dairy characteristics
	205	Animal qualified which has noticeable, but not predominant, dairy characteristics
106		Non-approved plant employees performing live animal identification
	206	Unidentified approved plant employee performing live animal identification
107		Carcass in cooler with CHB roll that does not have a "C" stamp or a USDA "Accepted as Specified" stamp over the "grade schedule" marking
	207	"C" stamp not placed on approved area of carcass
108		Identity of genetically qualified cattle is not maintained up to the point at which the "C" stamp is applied to the carcass
109		Carcass of a bull, bullock, or cow identified with "C" stamp

Defects 101, 201, 102, and 202 apply to cattle that are not genotype qualified. As long as no defects are observed under program monitoring the designated frequency of monitoring shall continue. However, observation of any major defect, two minor defects within a week, or three minor defects within four weeks, shall immediately result in accelerated monitoring. Under accelerated monitoring the frequency of monitoring shall be doubled. If no defects are observed during two weeks of accelerated monitoring, the plant shall revert to the normal monitoring level. If any major defect or two minor defects are observed under accelerated monitoring the identification program shall be placed under 100% MGCB supervision for four weeks. After four weeks of 100% supervision, the program will revert to accelerated monitoring before going to normal monitoring.

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THE EFFECT OF CATTLE GENOTYPE ON TEXTURE OF SELECTED MUSCLES DURING POST-MORTEM AGEING

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ABSTRACT

Changes in textural parameters of selected muscles (*m. longissimus lumborum* – LL, *m. semitendinosus* – ST, *m. semimembranosus* – SM, *m. biceps femoris* – BF) of pure-breed cattle Charolaise (CHL) and two groups of cross breeds: Hereford×Charolaise (HEF×CHL) and Simental×Charolaise (SIM×CHL) taken on the 3rd, 7th and 12th day of post-mortem cold storage were estimated.

Of the four muscles tested, the highest hardness, springiness and chewiness were found in BF, which at the same time showed the highest stringiness, perceptible of connective tissue, cooking loss, and the lowest juiciness. Post mortem ageing resulted in a reduction of hardness, springiness and cooking loss and in an increase of tenderness and purge loss. The rate and dimension of tenderization were relative to the both cattle genotype and muscle tested on the one side, and stage of tenderization on the other.

Key words: texture, muscles, young bulls, ageing

INTRODUCTION

The eating quality of beef (*i.a.* texture, tenderness, chemical composition) is related to the both ante- and post-mortem factors. Of the ante-mortem factors tested, the most important is an animal breed selection, which could be connected, for example, owing to cross-breeding programs, with meat quality [5]. A meat tenderness could have been achieved during post-mortem ageing. In case of cattle it is a long lasting period, and tenderization

process is estimated to the end even 2 weeks after slaughter. Post-mortem ageing of meat is connected with a proteolysis of myofibrils and connective tissue [8, 15, 22].

Proteolytic degradation of there proteins would cause texture changes, and, thus tenderization. However, the rate of tenderization could be correlated with many factors *i.a.* with a cattle breed and the muscle tested. The different susceptibility of the muscles or meat of different animal breeds could have been caused by *i.a.* different contents of connective tissue, and an activity of proteolytic enzymes – calpain and calpastatin on the one hand [25], different contents of red and white fibres in the muscles under consideration on the other [24]. It can be thus assumed that the properly genetic selection in beef production will allow to obtain meat of high eating quality (tenderness).

AIM OF THE STUDY

The study presented here was aimed at comparing changes of texture of selected muscles from Charolaise pure breed cattle and their crosses with Hereford and Simental breeds during post-mortem ageing.

MATERIAL AND METHODS

The study involved 3 groups of cattle of following genotypes:

1. group: ♀ CHL x ♂ CHL
2. group: ♀ HEF x ♂ CHL
3. group: ♀ SIM x ♂ CHL,

where: CHL – Charolaise breed, HEF-Hereford breed, SIM-Simental breed

Eight bulls of each group were examined. An investigation have been done at private stock-farm in Wyszobór. The young bulls were managed at their mothers in pasture system. All of them were born in early spring (March, April). Mother's milk and green forage from pasture (additionally, at night, a hay and protein food) was fed during a rearing period. During the fattening period the young bulls were allowed a fattening diet (a grass and maize ensilage with a small quantity of cereal mix – a barley, oat and wheat rye). Body weight gain of the animals, of about 850.g daily, were recorded. Animals were slaughtered at the mass of 550kg at the experimental station. The bulls were transported to the slaughterhouse of Meat Processing Plant of Agrofirma Witkowo, and then were starved by about 12 h prior to slaughtering. A left hindquarters of carcass, kept at the cold room for 12 h served to obtain 4 muscles:

- LL – *m. longissimus lumborum*,
- ST – *m. semitendinosus*,
- SM – *m. semimembranosus*,
- BF – *m. biceps femoris*.

About 4 cm thick slices were cut perpendicularly to the fibres from each muscle. Each slice, after weighing, was vacuum packaged in plastic bags and cold stored 4±1°C. The samples were taken for assays on the 3rd, 7th and 12th day of *post mortem (p.m.)* cold storage.

After removal of the plastic bags, the pH was measured with a pH-meter CP-215. After weighing, samples were tightly wrapped in thermoresistant plastic bags, and cooked in water at 80°C until the geometric centre of a sample was heated to 70°C. The cooked samples were cooled to 15°C, re-weighed, wrapped in plastic sheet and cold stored until the analysis were made. Before the texture assays were made, 20±1mm thick slices were cut from each muscle with an electric knife Siemens.

An instrumental texture assays
The texture was evaluated on an Instron 1140 apparatus, using the TPA double squeeze test. The test involved driving a 6.2 mm diameter shaft twice into a sample down to 80% of its height, pararell to the fibres; the force was applied at a speed of 50 mm/min. The force-deformation curve obtained served to calculate meat hardness, cohesiveness, springiness and chewiness [2]. The procedure was repeated 9 times on each sample batch.

The sensory texture evaluation

The sensory evaluation of the beef samples was assessed by a trained expert panel of 4 members with, in general, a minimum of four years experience in texture analysis of meat and meat products. The meat tenderness, juiciness, perceptible of connective tissue and stringiness were assessed using a 5-points scale (Table 1).

Table 1. The 5-points sensory evaluation scale

Traits	1 point	2 points	3 points	4 points	5 points
Tenderness	the toughest	tough	average tough	tender	the most tender
Juiciness	the most dry	dry	average dry	juicy	the most juicy
Perceptible of connective tissue	very abundant	easy perceptible	average perceptible	weakly perceptible	imperceptible
Stringiness	the most stringy	easy perceptible	average perceptible	weakly perceptible	imperceptible

The calculation of purge and cooking losses

Purge loss (%) was calculated from the difference in weight before and after post-mortem ageing. Cooking loss (%) was calculated from the difference in weight before and after thermal treatment.

Statistical treatment

All the calculations were performed with Statistica v.6.0 PL software. A mean and standard deviation was calculated for each samples. A significance of differences was explored by applying Student's *t* test at $P = 0.05$.

RESULT AND DISCUSSION

Table 2 presents the textural parameters of four beef muscles (of the three cattle groups) after 3rd, 7th and 12th days of post-mortem ageing. A comparison between textural parameters of beef (an arithmetical means of 4 muscles) showed no significant differences in TPA parameters between 3 groups of cattle in any post-mortem ageing period tested. However, the muscle samples of Charolaise pure-breed taken on the 3rd and 7th day ageing showed the highest hardness, cohesiveness, springiness and chewiness. Whereas, meat of SIM×CHL cross-breeds was the toughest, the most cohesiveness, springy and chewy after 12th days of ageing. The sensory texture evaluation showed no significant differences in parameters tested between groups of cattle (Table 3). Among the 3 groups of animals, of the all ageing period tested, CHL meat was characterized by the highest tenderness, the highest juiciness being recorded in HEF×CHL crosses meat, muscles of SIM×CHL showed the lower degree of connective tissue perceptible. The technological properties (pH, purge and cooking losses) showed no significant differences between meat of bulls tested, too (Table 4). Regardless of the ageing time, the highest pH values, and cooking loss were recorded in the SIM×CHL crosses meat, the lowest values being typical of the HEF×CHL crosses muscles.

No differences in meat quality (*i.a.* texture, technological properties) of different cattle groups was reported also by Maher et al. [12], and Chambaz et al. [5]. While Monson et al. [14] showed the differences in hardness between cattle breeds only for LD muscle on 1st, 3rd, and 7th day post-mortem, and next on 14th day of ageing, no differences. The differences between results obtained by those authors and Monson et al. [14] could have been probably caused by differences in experimental materials. A higher differences in meat quality may have resulted from a comparison of pure-breeds [14, 17] than during a comparison pure-breed cattle with their crosses [13] as well as a comparison between cattle types of the same breed [12].

According to Belew et al. [1] and Maher et al. [12], the differences in eating quality of beef may have resulted from differences between breeds, as well as between genotype groups or within an animal carcass. Numerous authors [1, 3, 6, 17] found bovine muscle to differ in terms of hardness. The results of the analyses performed show that of the four muscles tested, regardless of the cattle genotype on the one hand, and ageing time on the other, the highest hardness, springiness, chewiness and stringiness were typical of the BF (Table 2), which at the same time was characterized by the lowest juiciness, the highest perceptible of connective tissue (Table 3) and purge and cooking losses (Table 4). Whereas LL, ST and SM muscles were intermediate in terms of their textural and technological properties. A similar texture ordering of animals muscles was reported by Wheeler et al. [26, 27], and Torrescano et al. [23] for cattle, Sobczak et al. [21], Lachowicz et al. [10] for pigs muscles, whereas for wild boars muscles by Lachowicz et al. [11].

Table 2. Mean values of textural parameters of 4 bovine muscles according to cattle genotype and ageing period

	♀ CHL x ♂ CHL					♀ HEF x ♂ CHL					♀ SIM x ♂ CHL				
	LL	ST	SM	BF	$\bar{x}$	LL	ST	SM	BF	$\bar{x}$	LL	ST	SM	BF	$\bar{x}$
Samples after 3 days of ageing															
Hardness (N)	60.22^{a}_{1}	63.12^{ab}_{1}	70.52^{b}_{1}	89.81^{c}_{1}	***70.92***	61.74^{a}_{1}	64.66^{a}_{1}	70.10^{b}_{1}	72.58^{b}_{2}	***67.27***	67.56^{ab}_{1}	64.19^{a}_{1}	66.16^{ab}_{1}	71.54^{b}_{2}	***67.36***
Cohesiveness (-)	0.516^{ab}_{1}	0.546^{b}_{1}	0.517^{ab}_{1}	0.476^{c}_{1}	***0.514***	0.444^{a}_{2}	0.473^{ab}_{2}	0.430^{a}_{2}	0.531^{b}_{1}	***0.470***	0.520^{a}_{1}	0.490	0.490^{a}_{1}	0.480^{a}_{1}	***0.495***
Springiness (cm)	1.19^{a}_{1}	1.28^{b}_{1}	1.25^{ab}_{1}	1.40^{c}_{1}	***1.28***	1.10^{a}_{1}	1.20^{b}_{1}	1.16^{ab}_{1}	1.30^{c}_{2}	***1.19***	1.14^{a}_{1}	1.32^{b}_{1}	1.26^{b}_{1}	1.34^{b}_{2}	***1.26***
Chewiness (N×cm)	37.29^{a}_{1}	44.11^{b}_{1}	45.22^{b}_{1}	60.49^{c}_{1}	***46.78***	30.15^{a}_{2}	36.70^{a}_{2}	35.71^{a}_{2}	50.09^{b}_{2}	***38.16***	40.54^{a}_{1}	41.02^{a}_{1}	40.85^{a}_{1}	46.01^{a}_{2}	***42.10***
Samples after 7 days of ageing															
Hardness (N)	53.94^{a}_{1}	58.22^{a}_{1}	58.54^{a}_{1}	81.03^{b}_{1}	***62.93***	55.94^{a}_{1}	58.59^{a}_{1}	56.53^{a}_{1}	69.95^{b}_{2}	***60.25***	59.73^{a}_{1}	61.45^{a}_{1}	57.98^{a}_{1}	65.21^{a}_{2}	***61.09***
Cohesiveness (-)	0.506^{a}_{1}	0.524^{a}_{1}	0.456^{b}_{1}	0.506^{a}_{1}	***0.498***	0.453^{a}_{2}	0.541^{b}_{1}	0.520^{b}_{2}	0.428^{a}_{2}	***0.486***	0.494^{a}_{1}	0.539^{a}_{1}	0.535^{a}_{2}	0.506^{a}_{1}	***0.518***
Springiness (cm)	1.10^{a}_{1}	1.23^{b}_{1}	1.23^{b}_{1}	1.22^{b}_{1}	***1.20***	1.06^{a}_{1}	1.18^{bc}_{1}	1.14^{b}_{2}	1.26^{c}_{1}	***1.16***	1.11^{a}_{1}	1.28^{b}_{2}	1.24^{b}_{1}	1.24^{b}_{1}	***1.22***
Chewiness (N×cm)	29.63^{a}_{1}	37.40^{b}_{1}	32.68^{ab}_{1}	50.02^{c}_{1}	***37.43***	29.98^{a}_{1}	37.40^{b}_{1}	33.68^{ab}_{1}	37.72^{b}_{2}	***34.70***	30.00^{a}_{1}	42.27^{b}_{1}	38.46^{b}_{1}	40.92^{b}_{2}	***37.91***
Samples after 12 days of ageing															
Hardness (N)	52.64^{a}_{1}	52.48^{a}_{1}	50.54^{a}_{1}	68.32^{b}_{1}	***56.00***	55.23^{a}_{1}	54.66^{a}_{1}	51.89^{a}_{1}	58.61^{a}_{2}	***55.10***	58.52^{a}_{1}	55.36^{a}_{1}	52.20^{a}_{1}	$61.26^{a}_{1,2}$	***56.84***
Cohesiveness (-)	0.454^{a}_{1}	0.462^{a}_{1}	0.508^{b}_{1}	0.432^{a}_{1}	***0.464***	0.441^{a}_{1}	0.474^{a}_{1}	0.499^{a}_{1}	0.480^{a}_{1}	***0.474***	0.475^{ab}_{1}	0.524^{b}_{2}	0.437^{a}_{2}	0.473^{ab}_{1}	***0.477***
Springiness (cm)	1.03^{a}_{1}	1.22^{b}_{1}	1.07^{a}_{1}	1.16^{b}_{1}	***1.12***	1.05^{a}_{1}	1.16^{b}_{1}	1.01^{a}_{1}	1.14^{b}_{1}	***1.09***	1.06^{a}_{1}	1.24^{b}_{1}	1.17^{b}_{2}	1.17^{b}_{1}	***1.16***
Chewiness (N×cm)	25.22^{a}_{1}	29.58^{ab}_{1}	27.63^{a}_{1}	33.84^{b}_{1}	***27.07***	25.57^{a}_{1}	30.05^{ab}_{1}	26.15^{a}_{1}	32.07^{b}_{1}	***28.46***	28.97^{ab}_{1}	35.97^{c}_{1}	26.69^{a}_{1}	33.90^{bc}_{1}	***31.38***

a, b - samples in a rows, denoted by different letters, were significantly different within a breed ($P \leq 0.05$);
1,2 - samples in a rows, denoted by different numerals, were significantly different between breeds ($P \leq 0.05$)

Table 3. Mean values of sensory attributes of 4 bovine muscles according to cattle genotype and ageing period

	♀ CHL x ♂ CHL					♀ HEF x ♂ CHL					♀ SIM x ♂ CHL				
	LL	ST	SM	BF	$\overline{X}$	LL	ST	SM	BF	$\overline{X}$	LL	ST	SM	BF	$\overline{X}$
Samples after 3 days of ageing															
Tenderness (points)	2.4^{a}_{1}	3.5^{b}_{1}	3.2^{b}_{1}	2.2^{a}_{1}	***2.82***	2.6^{a}_{1}	3.0^{a}_{1}	3.0^{a}_{1}	2.5^{a}_{1}	***2.78***	2.8^{a}_{1}	3.0^{a}_{1}	2.7^{a}_{1}	2.3^{a}_{1}	***2.70***
Juiciness (points)	3.2^{a}_{1}	4.0^{b}_{1}	2.2^{c}_{1}	2.2^{c}_{1}	***2.90***	3.2^{a}_{1}	2.5	2.5^{b}_{1}	3.5^{a}_{2}	***2.92***	2.7^{a}_{1}	2.8^{a}_{2}	2.7^{a}_{1}	2.5^{a}_{1}	***2.68***
Perceptible of connective tissue (points)	3.2^{a}_{1}	3.0^{a}_{1}	2.5^{a}_{1}	2.5^{a}_{1}	***2.80***	3.5^{a}_{1}	2.5^{b}_{1}	2.8^{b}_{1}	2.8^{b}_{1}	***2.90***	2.8^{a}_{1}	2.5^{a}_{1}	2.5^{a}_{1}	2.0^{a}_{1}	***2.45***
Stringiness (points)	3.2^{a}_{1}	3.0^{a}_{1}	3.0^{a}_{1}	3.0^{a}_{1}	***3.05***	3.8^{a}_{2}	3.0^{b}_{1}	2.5^{b}_{1}	3.0^{b}_{1}	***3.08***	2.8^{a}_{1}	3.0^{a}_{1}	3.2^{a}_{1}	2.7^{a}_{1}	***2.92***
Samples after 7 days of ageing															
Tenderness (points)	2.8^{a}_{1}	3.8^{b}_{1}	3.5^{b}_{1}	2.4^{a}_{1}	***3.12***	2.7^{a}_{1}	3.8^{b}_{1}	3.1^{a}_{1}	2.8^{a}_{1}	***3.10***	3.0^{a}_{1}	3.3^{a}_{1}	3.0^{a}_{1}	2.5^{a}_{1}	***2.95***
Juiciness (points)	2.2^{a}_{1}	3.8^{bc}_{1}	2.8^{ac}_{1}	3.2^{c}_{1}	***3.00***	3.8^{a}_{2}	3.0^{b}_{2}	2.8^{b}_{1}	4.0^{a}_{2}	***3.40***	3.0^{a}_{1}	$3.3^{a}_{1,2}$	2.8^{a}_{1}	3.3^{a}_{1}	***3.10***
Perceptible of connective tissue (points)	2.2^{a}_{1}	3.5^{b}_{1}	3.0^{b}_{1}	3.0^{b}_{1}	***2.92***	2.5^{a}_{1}	$3.0^{a}_{1,2}$	2.8^{a}_{1}	2.2^{a}_{2}	***2.62***	3.7^{a}_{2}	2.7^{b}_{2}	3.0^{b}_{1}	3.5^{ab}_{1}	***3.22***
Stringiness (points)	3.0^{a}_{1}	3.2^{a}_{1}	2.5^{a}_{1}	3.0^{a}_{1}	***2.92***	3.2^{a}_{1}	3.2^{a}_{1}	2.8^{a}_{1}	3.5^{a}_{1}	***3.18***	3.3^{a}_{1}	3.3^{a}_{1}	3.2^{a}_{1}	3.0^{a}_{1}	***3.20***
Samples after 12 days of ageing															
Tenderness (points)	3.2^{a}_{1}	4.0^{b}_{1}	4.0^{b}_{1}	2.6^{a}_{1}	***3.45***	3.1^{a}_{1}	4.0^{b}_{1}	3.2^{a}_{2}	3.0^{a}_{2}	***3.32***	3.2^{a}_{1}	3.3^{a}_{2}	3.2	2.6^{a}_{1}	***3.08***
Juiciness (points)	3.0^{a}_{1}	3.0^{a}_{1}	$3.5^{a}_{1,2}$	3.0^{a}_{1}	***3.12***	3.0^{a}_{1}	3.5^{ab}_{1}	4.0^{b}_{2}	3.0^{a}_{1}	***3.38***	3.2^{a}_{1}	3.0^{a}_{1}	3.3^{a}_{1}	2.8^{a}_{1}	***3.08***
Perceptible of connective tissue (points)	$2.0^{a}_{1,2}$	3.5^{b}_{1}	3.5^{b}_{1}	2.5^{a}_{1}	***2.88***	2.5^{a}_{2}	3.5^{b}_{1}	4.0^{b}_{1}	3.5^{b}_{2}	***3.38***	2.7^{a}_{2}	2.8^{a}_{2}	2.8^{a}_{2}	2.5^{a}_{1}	***2.70***
Stringiness (points)	3.0^{a}_{1}	4.0^{b}_{1}	2.0^{c}_{1}	2.5^{ac}_{1}	***2.88***	3.0^{a}_{1}	$3.5^{a}_{1,2}$	3.5^{a}_{2}	3.0^{a}_{1}	***3.25***	3.2^{a}_{1}	3.0^{a}_{2}	3.3^{a}_{2}	3.2^{a}_{1}	***3.18***

a, b - samples in a rows, denoted by different letters, were significantly different within a breed ($P \leq 0.05$);
1,2 - samples in a rows, denoted by different numerals, were significantly different between breeds ($P \leq 0.05$)

Table 4. Mean values of pH, purge and cooking loss of 4 bovine muscles according to cattle genotype and ageing period

	♀ CHL x ♂ CHL					♀ HEF x ♂ CHL					♀ SIM x ♂ CHL				
	LL	ST	SM	BF	$\overline{X}$	LL	ST	SM	BF	$\overline{X}$	LL	ST	SM	BF	$\overline{X}$
Samples after 3 days of ageing															
pH	5.57^{a}_{1}	5.60^{a}_{1}	5.54^{a}_{1}	5.54^{a}_{1}	***5.56***	5.58^{a}_{1}	5.52^{a}_{1}	5.49^{a}_{1}	5.51^{a}_{1}	***5.52***	5.65^{a}_{1}	5.63^{a}_{1}	5.55^{a}_{1}	5.57^{a}_{1}	***5.60***
Purge loss (%)	1.74^{a}_{1}	1.97^{a}_{1}	2.16^{a}_{1}	2.64^{a}_{1}	***2.13***	1.94^{a}_{1}	2.68^{a}_{1}	2.48^{a}_{1}	2.92^{a}_{1}	***2.51***	1.41^{a}_{1}	1.78^{a}_{1}	1.98^{a}_{1}	2.56^{a}_{1}	***1.93***
Cooking loss (%)	22.70^{a}_{1}	28.92^{ab}_{1}	28.05^{ab}_{1}	33.73^{b}_{1}	***28.35***	24.30^{a}_{1}	28.25^{ab}_{1}	29.80^{ab}_{1}	31.15^{b}_{1}	***28.38***	25.39^{a}_{1}	29.49^{ab}_{1}	30.16^{b}_{1}	30.59^{b}_{1}	***28.91***
Samples after 7 days of ageing															
pH	5.54^{a}_{1}	5.56^{a}_{1}	5.55^{a}_{1}	5.54^{a}_{1}	***5.55***	5.58^{a}_{1}	5.52^{a}_{1}	5.52^{a}_{1}	5.51^{a}_{1}	***5.53***	5.64^{a}_{1}	5.63^{a}_{1}	5.67^{a}_{1}	5.57^{a}_{1}	***5.63***
Purge loss (%)	2.28^{a}_{1}	3.14^{a}_{1}	2.95^{a}_{1}	3.22^{a}_{1}	***2.90***	3.42^{a}_{1}	3.42^{a}_{1}	3.80^{a}_{1}	3.01^{a}_{1}	***3.41***	3.26^{a}_{1}	3.72^{a}_{1}	4.40^{a}_{1}	3.31^{a}_{1}	***3.67***
Cooking loss (%)	22.44^{a}_{1}	$26.8^{ab}_{1}0$	26.96^{ab}_{1}	30.44^{b}_{1}	***26.66***	22.80^{a}_{1}	26.40^{ab}_{1}	28.20^{ab}_{1}	30.60^{b}_{1}	***27.00***	22.63^{a}_{1}	27.99^{ab}_{1}	27.38^{b}_{1}	28.90^{b}_{1}	***26.73***
Samples after 12 days of ageing															
pH	5.60^{a}_{1}	5.61^{a}_{1}	5.60^{a}_{1}	5.56^{a}_{1}	***5.59***	5.54^{a}_{1}	5.56^{a}_{1}	5.54^{a}_{1}	5.54^{a}_{1}	***5.54***	5.60^{a}_{1}	5.63^{a}_{1}	5.61^{a}_{1}	5.57^{a}_{1}	***5.60***
Purge loss (%)	4.00^{a}_{1}	4.87^{a}_{1}	3.44^{a}_{1}	3.73^{a}_{1}	***4.01***	4.65^{a}_{1}	4.66^{a}_{1}	4.50^{a}_{1}	4.04^{a}_{1}	***4.46***	3.72^{a}_{1}	5.13^{b}_{1}	4.78^{ab}_{1}	3.98^{a}_{1}	***4.40***
Cooking loss (%)	20.71^{a}_{1}	20.30^{a}_{1}	22.53^{a}_{1}	24.12^{a}_{1}	***21.92***	18.40^{a}_{1}	19.40^{a}_{1}	21.85^{a}_{1}	28.83^{b}_{1}	***22.12***	21.21^{a}_{1}	25.35^{a}_{1}	26.44^{a}_{1}	27.15^{a}_{1}	***25.04***

a, b - samples in a rows, denoted by different letters, were significantly different within a breed ($P \leq 0.05$);
1,2 - samples in a rows, denoted by different numerals, were significantly different between breeds ($P \leq 0.05$)

According to Koohmaraie et al. [9], sarcomere length, connective tissue content, and proteolysis of myofibrillar proteins account for most of the explainable variation observed in tenderness of aged meat, after post-mortem storage. For example, while sarcomere length is the major determinant of PM muscle tenderness, connective tissue content is a major contributor to tenderness of BF muscle and proteolysis is the major determinant of L tenderness. Therefore, a similar hardness of LL, ST and SM muscles may have resulted from *i.a.* no differences in amounts and thickness of connective tissue in muscles. As shown by Brooks and Savell [3] muscles with thicker perimysium had a higher shear force. According to Wheeler et al. [26], BF is characterized by the most abundant connective tissue compared to other muscles tested – which at the same time, as shown in this work, was the toughest muscle; the lower amount of connective tissue being typical of the SM muscle, and the lowest was recorded in the LL.

The prolonged storage of meat from 3-7 days (the first stage of ageing), and the next from 7-12 days (the second stage of ageing) caused a decrease of hardness, springiness, chewiness and cooking losses and an increase in tenderness, juiciness and purge loss. The differences in mean values of parameters tested changes were dependent on both cattle genotype and kind of muscle on the one hand, and the stage of ageing on the other. No significant effects of ageing time on cohesiveness, perceptible of connective tissue, stringiness and pH changes were found.

Among the animal groups tested, the higher hardness, springiness and chewiness changes of CHL pure-breed meat were found at the first (between 3-7 days of cold storage) and the second (between 7-12 days) stage of ageing. The meat of cross-breeds animals was characterized by the higher susceptibility to tenderness, juiciness, purge and cooking loss changes during first stage of ageing, whereas CHL meat was more susceptible to changes at second stage of ageing.

Regardless of the cattle group, of all the muscles tested, the most susceptible to hardness, springiness and chewiness changes were BF and SM. However the higher changes of those parameters values were observed in SM at the first stage of ageing, and at the second stage in BF. The lowest hardness and chewiness changes were recorded in the LL, whereas the lowest springiness changes being typical of the ST. No effects of muscle on sensory parameters, as well as purge and cooking losses changes during ageing were found.

A similar effect of ageing time on muscle texture was reported by *i.a.* Koohmaraie et al. [9], Kołczak et al. [7], Palka [18], Brooks and Savell [3], Maher et al. [12], Monson et al. [14]. The data obtained show the rate and dimension of tenderization were dependent on muscle type, cattle genotype and ageing time. A comparison between the dimension of tenderization of four muscles showed the harder muscles (BF and SM) were the most susceptible to hardness changes. Kołczak et al. [7] demonstrated that the lower rate of tenderization being typical of the most tender muscle – PM, compared to harder ST muscle, whereas Hwang et al. [6] found that LD was characterized by reduced post-mortem proteolysis compared to ST muscle.

Of the cattle groups tested, the muscles of pure-breed bulls showed initially the highest hardness (at 3rd day post-mortem), and were the most susceptible to hardness changes; it is conformable to Sañudo et al. [20] and Monson et al. [14] research, who observed the bovine muscles of higher hardness to be more susceptible to hardness changes during ageing. Moreover, according Maher et al. [12], meat from double-muscle Charolaise cattle with higher hardness become more tender during first stage of ageing (2-7 days), while the cattle with normal muscles during second stage (between 7th and 12th day) of ageing. However, Monson et al. [14] is of the opinion that the meat from cattle of initially lower hardness is more susceptible to tenderization during first stage of ageing, while during the second stage more susceptible are breeds with muscles of higher hardness, but no significant differences between bovine muscles susceptible to hardness reduction during 21st and 35th day of ageing were found. According to Campo et al. [4], the animal breed is the minor determinant to tenderness of muscles, and the major contributor to tenderness is the ageing time. As shown in this work, regardless of both cattle genotype and muscle type, a higher hardness and tenderness changes were found between 3rd and 7th day (the first stage) than between 7th and 12th day (the second stage) of ageing. Similar observation were reported by Koohmaraie et al. [9], who observed the highest decrease of shear force between 1st and 3rd day post-mortem, while the reduction of shear force was insignificant between 3rd and 14th day of ageing. However Ruiz de Huidobro et al. [19] found no correlation between ageing time and drip loss, shear force or texture of L muscle, but a decrease at first stage of ageing and an increase at second stage of those parameters were observed.

CONCLUSIONS

The result obtained show no significant differences in meat eating quality between the three groups of cattle tested. However, the pure-breed CHL cattle meat, compared to the HEF×CHL and SIM×CHL crosses were characterized by the insignificant higher hardness, springiness and chewiness. Regardless of the young bulls genotype, among the four muscle tested, the highest values of hardness, springiness and chewiness were recorded in BF, which at the same time showed the lower, juiciness as well as the higher stringiness and the highest perceptible of connective tissue and purge and cooking losses. No significant differences in textural parameters and technological properties between LL, ST and SM were found.

Post-mortem ageing resulted in a reduction of muscle hardness, springiness, cooking loss and an increase of tenderness and purge loss, however the rate and dimension of tenderization were dependent on cattle genotype, muscle type and ageing time. Of the cattle groups tested, the muscles of CHL pure-breed bulls were the most susceptible to tenderization, and of the muscles tested – BF and SM. As shown in this work, a higher hardness changes were found at the first stage than at the second stage of ageing. An ageing process, as a result of a higher susceptibility on tenderization of muscles with a higher initially hardness, caused a decrease in hardness differences between muscles tested on the one hand, and between groups of cattle on the other.

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